UNITED STATES DEPARTMENT OF COMMERCE
Patent and Trademark Offic
Addr ss: COMMISSIONER OF PATENTS AND TRADEMARKS
Washington, D.C. 20231

| APPLICATION NO. | FILING DATE | FIRST NAMED INVENTOR | ATTORNEY DOCKET NO. |
|---|---|---|---|
| 09/165,088 | 09/30/98 | RAO M | T5550.CIP |

020450
ALAN J HOWARTH
PO BOX 1909
SANDY UT 84091

HM32/1005

| EXAMINER |
|---|
| KERR, J |

| ART UNIT | PAPER NUMBER |
|---|---|
| 1655 | |

DATE MAILED: 10/05/99

**Please find below and/or attached an Office communication concerning this application or proceeding.**

**Commissioner of Patents and Trademarks**

# Office Action Summary

| Application No. | Applicant(s) |
|---|---|
| 09/109,858 | Rao et al. |
| **Examiner** | **Group Art Unit** |
| Janet M. Kerr | 1633 |

☒ Responsive to communication(s) filed on *Jul 2, 1998* .

☐ This action is **FINAL.**

☐ Since this application is in condition for allowance except for formal matters, **prosecution as to the merits is closed** in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11; 453 O.G. 213.

A shortened statutory period for response to this action is set to expire *3* month(s), or thirty days, whichever is longer, from the mailing date of this communication. Failure to respond within the period for response will cause the application to become abandoned. (35 U.S.C. § 133). Extensions of time may be obtained under the provisions of 37 CFR 1.136(a).

**Disposition of Claims**

☒ Claim(s) *1-59* is/are pending in the application.

Of the above, claim(s) _____ is/are withdrawn from consideration.

☐ Claim(s) _____ is/are allowed.

☒ Claim(s) *1-59* is/are rejected.

☐ Claim(s) _____ is/are objected to.

☐ Claims _____ are subject to restriction or election requirement.

**Application Papers**

☐ See the attached Notice of Draftsperson's Patent Drawing Review, PTO-948.

☐ The drawing(s) filed on _____ is/are objected to by the Examiner.

☐ The proposed drawing correction, filed on _____ is ☐approved ☐disapproved.

☐ The specification is objected to by the Examiner.

☐ The oath or declaration is objected to by the Examiner.

**Priority under 35 U.S.C. § 119**

☐ Acknowledgement is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d).

☐ All ☐ Some* ☐ None of the CERTIFIED copies of the priority documents have been

☐ received.

☐ received in Application No. (Series Code/Serial Number) _____ .

☐ received in this national stage application from the International Bureau (PCT Rule 17.2(a)).

*Certified copies not received: _____ .

☐ Acknowledgement is made of a claim for domestic priority under 35 U.S.C. § 119(e).

**Attachment(s)**

☒ Notice of References Cited, PTO-892

☒ Information Disclosure Statement(s), PTO-1449, Paper No(s). *4*

☐ Interview Summary, PTO-413

☐ Notice of Draftsperson's Patent Drawing Review, PTO-948

☐ Notice of Informal Patent Application, PTO-152

*--- SEE OFFICE ACTION ON THE FOLLOWING PAGES ---*

## DETAILED ACTION

The Information Disclosure Statement, filed on 2/1/99, has been entered.

Claims 1-59 are being examined on the merits.

### *Claim Objections*

Claims 38-40 are objected to under 37 CFR 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicant is required to cancel the claim(s), or amend the claim(s) to place the claim(s) in proper dependent form, or rewrite the claim(s) in independent form. Claims 38-40 are directed to a method. However, the claims are dependent on claim 34 which is directed to a composition. The methods of claims 38-40 do not further limit the composition of claim 34 and claims 38-40 are thus improperly dependent. To expedite prosecution, claims 38-40 will be assumed to depend on claim 37, which is directed to a method for treating a neuronal disorder in a mammal comprising administering the pharmaceutical composition of claim 35. Claim 44 is also objected to as the "or" should be changed to "for".

### *Claim Rejections - 35 USC § 112*

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 11, 35-43, and 45-59 are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

Claims 35-43, and 45-58 are directed to a method for treating a neurological or neurodegenerative disease comprising administering and effective amount of neuronal restricted

precursors cells or derivatives thereof or mixtures thereof, and pharmaceutical compositions comprising neuronal restricted precursor cells.

The specification is non-enabling for the claim designated method as the specification does not provide guidance as to how to identify all neurological or neurodegenerative diseases which can be successfully treated with neuronal restricted precursor cells, how to determine effective amounts of the cells for all neurological or neurodegenerative diseases, and how to target the cells to appropriate sites such that the cells are effective in treating the neurological or neurodegenerative disease.

The specification broadly discloses neuronal disorders which can be treated by cell therapy or *ex vivo* gene therapy strategies. However, the specification is non-enabling for the claimed methods as the specification does not provide sufficient guidance as to how one of ordinary skill in the art would treat a patient having a neuronal disorder. The specification does not disclose any specific neuronal disorder which has been subjected to the claim-designated treatment regimen, nor does the specification teach any specific methodology associated with such a treatment regimen including the number of cells to be administered for a specific neuronal disorder, the route of administration for a specific neuronal disorder, or the relevant cell therapy target site for the specific neuronal disorder. It should also be noted that the state of the art at the time of filing suggests that neuronal transplantation techniques and *in vivo* therapeutic effectiveness have not been established such that utilizing cells to treat neuronal disorders is routine or predictable. For example, Sanberg *et al.* (Nucleic Acids Symp. Ser., 38:139-142, 1998) disclose that "[p]erhaps the most serious problem faced in the field of cell transplantation is that of a host generated immune response to the grafted tissue. The prevailing strategy is to systemically immunosuppress the transplanted patient for extended periods of time. This, however, puts the patient at risk for other health problems" (see page 140, under "Issues of Graft Rejection"). Sanberg *et al.* also indicate that cell transplantation has also been used to treat diseases or conditions in which neurons die, such as stroke or Huntington's Disease. As these disorders involve multiple neuron populations and extensive cell death throughout the brain, it is

more difficult to treat these conditions using cell transplantation (see page 141, under "Cell Transplantation in Huntington's Disease).

Sabate *et al.* (Clinical Neuroscience, 3:317-321, 1996) indicate that while neurotrophic factors have been shown to promote the survival of particular neuronal populations, the use of classical pharmacotherapy for neurological diseases is restricted by constraints specific to the nervous system. In particular, the blood-brain barrier prevents access to the brain of numerous macromolecules of therapeutic value. Delivery of such molecules requires intracerebral or intracerebroventricular injection, and infusion using osmotic pumps when long-term treatments are necessary. Therefore, the combination of infectious risks and constraints of the delivery technique have precluded the generalized use of drugs (see page 317, right column, last paragraph, bridging page 318). Sabate *et al.* indicate that gene therapy should enable neurologists to overcome the problems raised by pharmacotherapy and grafting of embryonic cells. Gene transfer into a give brain structure should permit local and controlled expression and release of therapeutic products. Moreover, limiting the expression of trophic factors to the therapeutic target only should avoid the severe side effects associated with intravenous or ICV administration (see page 318, left column, first full paragraph). However, Sabate *et al.* caution that there are several important issues to be resolved if gene therapy for neurological diseases is to become a reality including (1) extent of transgene expression, (2) stability of transgene expression, (3) targeting of the cells, (4) safety of the procedure, and (5) the vector large-scale production capacity (see page 318, left column, under "Recombinant Adenovirus For Gene Therapy"). Sabate *et al.* indicate that the expression of adenoviral vectors persists for several months, possibly because the CNS is partially sheltered from the immune system. However, there has been a report that IC injection of a first-generation recombinant adenovirus into the rat striatum leads to an inflammatory response. Retrograde transport of adenovirus to neurons of the substantia nigra was associated with a delayed and less intense inflammation at this distant site. However, there was no immune response, allowing the vector to persist and be expressed for at least two months with little inflammation. This was not the case if the rats were administered a

secondary subcutaneous injection of adenovirus vector. This immunization led within 2 weeks to severe mononuclear inflammation and microglial activation in the striatum, and consequently local demyelination and a decrease in vector-encoded protein expression (see page 320, middle column, under "Future Developments For Adenovirus", bridging right column).

Scheffler *et al.* (TINS, 22:348-357, 1999) disclose that neural-transplantation paradigms must contend with the fact that, aside from rare exceptions, widespread neuron replacement is not easily accomplished in the normal postnatal and adult brain. Not only has the post-developmental genesis of neurons been presumed to be a rarity, but it is also generally assumed that as the brain matures after birth, it loses the ability to serve as a growth supportive host environment and, by contrast, is dominated by growth-inhibitory factors. Developmental cues might persist to certain degrees in the mature brain, but long-distance axonal growth and precise target seeking within established brain circuities are still troublesome. One transplant study of postnatal and adult dorsal-root-ganglion neurons into adult rat white matter tracts showed that grafted neurons had the ability to attempt such long-distance axonal growth, but it was the ECM-rich environment around a graft injection site that deterred most growth (see page 354, right column, second paragraph under "Transplantation and gene therapies").

With regard to *ex vivo* gene therapy, the specification does not teach a correlation between an effective treatment regimen and a neuronal disorder with the administration of neuronal restricted precursor cells transformed with the claim-designated genes. Similarly, the specification does not provide guidance as to the source of the nucleic acid to be used in the gene constructs, or suitable expression vectors which can be used in the transformation of the cells such that the cells express the protein of interest. Moreover, there is no teaching in the specification as to the effect of transforming the neuronal restricted precursor cells, or the effect of the expressed protein on the phenotypic characteristics of the neuronal restricted precursor cells, i.e., there is no teaching in the specification that the transformed cells would maintain morphologic and phenotypic characteristics which are required in the therapeutic effectiveness of the transformed cell population. In addition, there is no teaching in the specification of a

correlation between the gene to be expressed and the neuronal disorder, or whether the *ex vivo* gene therapy is effective in overcoming the neuronal disorder. It should also be noted that gene therapies for treating neuronal disorders are still problematic in view of the teachings of Sabate *et al.* as discussed above. Based on the lack of guidance in the specification, it would require undue experimentation to determine the appropriate constructs to be used in transforming neuronal restricted precursor cells such that these cells are effective in treating a neuronal disorder.

In view of the lack of guidance in the specification, the teachings in the prior art that cell and gene therapy strategies for treating neuronal disorders are not well established and are still problematic, and in view of the lack of guidance in the specification as to specific protocols in which the clinical efficacy of the claim-designated methods has been established, one of ordinary skill in the art would not be able to use the claim-designated methods or claim-designated cells predictably and without undue experimentation.

Claim 59 is drawn to a method of isolating a pure population of mammalian CNS neuron-restricted precursor cells which requires a sample of mammalian embryonic stem cells.

While the specification discloses mouse ES cells obtained from the Developmental Studies Hybridoma Bank (see page 55), there is no teaching in the specification as to a specific deposit number of the cells. Thus, it is unknown what these mouse ES cells are. Moreover, the specification does not provide guidance as to how to obtain embryonic stem cells from other species. At the time of filing, the state of the art is such that the generation of ES cells, i.e., cells which retain their totipotential capacity and are able to generate cells of all lineages, including germline, after being introduced into host a blastocyst, is neither routine nor predictable in species other than mice. Bradley (Biotechnology, 1992) teaches that while a number of reports have been made claiming isolation of ES cells from farm animals, the description of these cell lines is yet to be supported by the demonstration that they can proliferate and differentiate in an embryo, *in vivo*, contributing to somatic tissues or germ cells (see page 53, right column, last paragraph bridging page 54). Similarly, Seamark (Reproductive Fertility and Development, 1994) discloses

that totipotency for ES cell technology in many livestock species has not been demonstrated (page 6, Abstract). Moreover, Mullins *et al.* (J. Clin. Invest., 98:S37-S40, 1996) teach that while chimeric animals for several species has been produced using purported ES cells, germ line transmission of an ES cell has not been demonstrated in species other than mice. In view of the lack of guidance in the specification with regard to the process of obtaining mammalian embryonic stem cells, the use of the cells in the claimed cell culture method is not enabled.

Claims 1-59 are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for isolating a homogeneous population of a rat neuronal restricted progenitor cell, does not reasonably provide enablement for isolating a homogeneous population of any mammalian neuronal restricted progenitor cell. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make the invention commensurate in scope with these claims.

While the specification is enabling for the isolation and utilization of rat neuronal restricted precursor cells, the specification is non-enabling for the isolation and utilization of all mammalian neuronal restricted precursor cells. The specification teaches a distinct developmental time point in which rat neuronal restricted precursor cells can be isolated using specific antibodies. However, the specification does not provide information with respect to the appropriate developmental time points for other mammalian species, whether the reagent used in the isolation can be utilized for other mammalian species, or whether the morphologic and phenotypic properties of the neuronal restricted precursor cells obtained from the rat can be correlated with other mammalian species. Moreover, Svendsen *et al.* (TINS, 22:357-364, 1999) disclose that there are significant genetic differences between rodents and humans, reflected by the fact that many antibodies, probes and drugs are species-specific. Furthermore, it is now clear that human neural precursor cells are very different from their rodent counterparts and need to be assessed independently: it is not possible to extrapolate from mouse to man directly. Thus, based on the lack of guidance in the specification, and the teachings of Svendsen *et al.* that different species

need to be assessed independently, one of ordinary skill in the art would not be able to make or use the invention as claimed without undue experimentation.

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 1-59 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 1 is rendered vague and indefinite as it is unclear what morphologic and phenotypic characteristics are required to identify a "neuron-restricted precursor cell".

Claims 2, 3, and 8-12 are rendered vague and indefinite by the phrase "capable of" as it is unclear what phenotypic characteristics of the cells and what conditions are required such that the cells have the capability of self-renewal (claim 2), of differentiation (claims 3, 8, and 10), of releasing and responding to neurotransmitters (claim 8), of expressing neurotransmitter-synthesizing enzymes (claim 9), of stably expressing at least one material (claim 11), and of generating both neurons and glia (claim 12).

Claim 7 is rendered vague and indefinite by the phrase "mammalian embryo" as it is unclear at what developmental stage the embryo should be obtained for the method.

Claim 12 is rendered vague and indefinite for the following reasons: it is unclear from what source the population of mammalian multipotent CNS stem cells are obtained; how the cells are isolated; what the characteristics of the multipotent CNS stem cells are such that the cells can be identified as precursors/progenitors of neurons and glia; what ingredients are present in the medium such that the cells begin differentiating; how long would the cells incubate in the medium prior to the purification recited in step (c); what process steps are required for the purification of the cells in step (c); and it is unclear which selected antigen would suitably define the cells as

neuron-restricted precursor cells; and what type of medium and what culture conditions are required which support adherent cell growth. In addition, there appears to be a missing step in the method as the last step, i.e., step (d) is an incubation step, yet the preamble of the claim requires isolation of a pure population of mammalian CNS neuron-restricted precursor cells. At which point in the method is the pure population of mammalian CNS neuron-restricted precursor cells isolated?

Claim 17 is rendered vague and indefinite for the following reasons: it is unclear how to identify the CNS tissue at a developmental stage after closure of the neural tube but prior to differentiation of cells in the neural tube; it is unclear how are the neural tube cells are selectively dissociated from other cells in the CNS tissue; it is unclear what type of substratum is required to support adherent growth of neuroepithelial stem cells; it is unclear what are "effective amounts" of fibroblast growth factor and chick embryo extract which would support adherent growth of the neuroepithelial stem cells; and it is unclear what temperature and what type of atmosphere are conducive to growing neuroepithelial stem cells. In addition, the phrase "neuroepithelial stem cells" recited in step (c) lacks proper antecedent basis as there are no neuroepithelial stem cells recited in steps (a) or (b) of the claim.

Claim 20 is rendered vague and indefinite by the phrase "wherein said medium" as it is unclear if the intended medium is the medium recited in step (b), step (c), or both. The phrase lacks proper antecedent basis. In addition, it is unclear what is deemed "effective amounts" of fibroblast growth factor and neurotrophin 3 as it is unclear what the intended effects of the fibroblast growth factor and neurotrophin 3 are.

Claim 21 is rendered vague and indefinite for the following reasons: it is unclear if any portion of the CNS tissue can be used in the method; it is unclear which selected antigen, substratum, medium, temperature, and atmosphere are required for isolating the neuron-restricted precursor cells and supporting growth of the cells. The metes and bounds of the various elements are unclear. Moreover, the claim appears to be incomplete as there is no process step in which

the neuron-restricted precursor cells are recovered for isolating the pure population of neuron-restricted precursor cells as set forth in the preamble.

Claim 28 is rendered vague and indefinite for the following reasons: it is unclear how the cells are provided, and it is unclear what types of conditions are proliferating or differentiating conditions. Moreover, the claim appears to be incomplete as there is no process step in which the postmitotic neurons are recovered for obtaining postmitotic neurons as set forth in the preamble.

Claim 34 is rendered vague and indefinite by the phrase "neuron-restricted cells" as it is unclear if these are the same neuron-restricted precursor cells recited in claims 1-7 or if they are different cells. The phrase lacks proper antecedent basis.

Claim 35 is rendered vague and indefinite by the phrase "therapeutically effective amount" as there is no recitation of the type of therapy intended. Thus, it is unclear what amount of cells would be deemed therapeutically effective.

Claims 36 and 37 are rendered vague and indefinite as it is unclear how a neuronal disorder is defined, i.e., what are the identifying characteristics of the disorders which can be treated by the method? Moreover, it is unclear if the mode of "administering" is dependent on the disorder being treated and if the "therapeutically effective amount" is dependent on the disorder being treated.

Claim 39 is rendered vague and indefinite by the phrase "differentiation factors, growth factors, cell maturation factors" as it is unclear which factors allow the differentiation, growth and maturation of the neuronal restricted precursor cells. The metes and bounds of the phrase are unclear. Moreover, it is unclear what the distinction is between a differentiation factor and a cell maturation factor. Clarification is requested.

Claims 41 and 42 are rendered vague and indefinite for the following reasons: it is unclear how the cells are used as a delivery vehicle for delivery of an agent or trophic factor to glial cells or neurons, respectively, i.e., it is unclear what characteristics of the cells are required which render the cells capable of targeting glial cells and neurons. Moreover, with regard to claim 42, it

is unclear what factors are encompassed in "trophic factors". The metes and bounds of the phrase "trophic factors" are unclear.

Claim 43 is rendered vague and indefinite for the following reasons: it is unclear what type of neurodegenerative symptoms are intended, how the cells are genetically transformed, and whether the effective amount of cells to be administered is dependent on the symptom being treated.

Claim 44 is rendered vague and indefinite by the phrase "monitoring the reaction" as it is unclear what type of reactions should be monitored, e.g., proliferation, apoptosis, differentiation, and whether the monitoring is the same for all possible reactions.

Claim 45 is rendered vague and indefinite by the phrase "neurological or neurodegenerative disease" as it is unclear which diseases are intended and it is unclear if the effective amount of cells to be administered is dependent on the disease being treated.

Claims 46-48 are rendered vague and indefinite by the phrase "are caused to" as it is unclear how the cells are "caused to" proliferate and differentiate. Moreover, it is unclear how to differentiate the cells once they are administered *in vivo* as recited in claims 47 and 48.

Claims 44-49, 53, and 57 are rendered vague and indefinite by the term "derivative" as it is unclear what a derivative of a neuronal restricted precursor cell encompasses. Thus, the metes and bounds of the term are unclear.

Claim 58 is rendered vague and indefinite as it is unclear what gene, promoter, regulatory sequences, expression vector, etc. are required for making the genetically transduced neuronal restricted precursor cells. The metes and bounds of what is encompassed in a genetically transduced neuronal restricted precursor cell is unclear.

Claim 59 is rendered vague and indefinite for the following reasons: it is unclear which selected antigen, substratum, medium, temperature, and atmosphere are required for isolating the neuron-restricted precursor cells and supporting growth of the cells. The metes and bounds of the various elements are unclear.

The phrases "neuron-restricted precursor" and "neuronal restricted precursor" appear to be used interchangeably in the claims. It is unclear if the two phrases are indicative of one cell population or if the two phrases are identifying distinguishable cell populations. If the two phrases are indicative of the same cell population, it is requested that only one of the phrases be used throughout the claims for consistency. If the two phrases are identifying different cell populations, then clarification of the features associated with each cell population is requested.

### *Claim Rejections - 35 USC § 102*

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless --
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

Claims 1-10, 21-23, 25-27, and 34 are rejected under 35 U.S.C. 102(b) as being anticipated by Blass-Kampmann *et al.* (J. Neuroscience Research 37:359-373, 1994).

The claims are drawn to a composition containing a pure population of neuron-restricted precursor cells.

Blass-Kampmann *et al.* disclose a composition of rat Mab RB21-7 binding cells, i.e., cells which express embryonic neural cell adhesion molecule, isolated by FACS, and having a purity of 98-99%. The Mab RB21-7 binding cells, which are neural precursor cells, are capable of self-renewal (see page 364, under "Isolation...", "Phenotypic Properties...", and "Differentiation..."). In that embryonic neural cell adhesion molecule is a cell surface marker which is inherently expressed in neuron-restricted precursor cells, the neuron-restricted precursor cells of Blass-Kampmann *et al.* would inherently not differentiate into CNS glial cells, and would inherently not express a ganglioside recognized by A2B5 antibody (a marker of glia) or nestin (a marker of neuroepithelial cells). The cells of Blass-Kampmann *et al.*, therefore, have the same phenotypic and morphologic characteristics of the claimed cells. Moreover, inasmuch as neuron-restricted

precursors differentiate into neurons, the cells disclosed by Blass-Kampmann *et al.*, upon differentiation, would inherently release and respond to neurotransmitters, would inherently express neurotransmitter-synthesizing enzymes and would inherently form functional synapses and/or develop electrical activity absent evidence to the contrary.

In addition, Blass-Kampmann *et al.* disclose a method of dissociating cells from prenatal (prenatal days E12-F18) rat brains, sorting the brain cells by FACS analysis to obtain Mab RB21-7+ cells, and culturing the cells under different culturing conditions such as culturing the sorted cells in modified Eagle-Dulbecco medium supplemented with FCS on coverslips that had been successively treated with poly-L-ornithine and laminin, or in F-14 medium supplemented with FCS on coverslips coated with poly-L-lysine, and incubated at 37°C under a humidified atmosphere of 7.5% $CO_2$ in air (see pages 361-362). Inasmuch as there is no claim-designated isolation step of a homogeneous population of neuronal restricted precursor cells in the methods claims, the cell culture methods of Blass-Kampmann *et al.* anticipate the claimed cell culture methods.

Thus the cells and the methods disclosed by Blass-Kampmann *et al.* anticipate the claimed invention.

### *Claim Rejections - 35 USC § 103*

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor

and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(f) or (g) prior art under 35 U.S.C. 103(a).

Claims 12-20, 24, 28-33, and 44 are rejected under 35 U.S.C. 103(a) as being unpatentable over Blass-Kampmann *et al.* (J. Neuroscience Research 37:359-373, 1994), taken with Boss *et al.* (U.S. Patent No. 5,411,883, 1995), Weiss *et al.* (WO93/01275, 1993), Johe *et al.* (U.S. Patent No. 5,753,506, 1998, effective filing date of 5/23/96), Rao *et al.* (26th Annual Meeting of the Society for Neuroscience, 22:527, Abstract #215.12, 1996), and Lee *et al.* (U.S. Patent No. 5, 175, 103, 1992).

Blass-Kampmann *et al.* disclose a composition of rat Mab RB21-7 binding cells, i.e., cells which express embryonic neural cell adhesion molecule, isolated by FACS, and having a purity of 98-99%. The Mab RB21-7 binding cells, which are neural precursor cells, are capable of self-renewal (see page 364, under "Isolation...", "Phenotypic Properties...", and "Differentiation..."). In addition, Blass-Kampmann *et al.* disclose a method of dissociating cells from prenatal (prenatal days E12-F18) rat brains, sorting the brain cells by FACS analysis to obtain Mab RB21-7+ cells, and culturing the cells under different culturing conditions such as culturing the sorted cells in modified Eagle-Dulbecco medium supplemented with FCS on coverslips that had been successively treated with poly-L-ornithine and laminin, or in F-14 medium supplemented with FCS on coverslips coated with poly-L-lysine, and incubated at 37°C under a humidified atmosphere of 7.5% $CO_2$ in air (see pages 361-362). Depending on the prenatal age and the culture conditions, the isolated cells differentiate into neurons or glia. Blass-Kampmann *et al.* indicate that separation of FACS of E-N-CAM+ cells from whole brain cell suspensions and their cultivation *in vitro* revealed that the cellular composition at later stages of culture was dependent on the stage of development at the start of cultivation. For example, increasing proportions of astrocyte precursors were observed the later in development E-N-CAM+ cells were isolated. Most of the Mab RB21-7 binding cells isolated on prenatal days F15 and F18 differentiated into neuronal phenotypes in F-14 medium, expressing either Mab Q211 binding antigens or the p68

subunit of NF. E-N-CAM+ cells separated from prenatal day F22 brains showed no potential for further neuronal differentiation in both F-14 and modified Eagle-Dulbecco medium, and died after 24 hr. In contrast, Mab RB21-7+ cells in whole F22 brain cultures exhibited a well-differentiated neuronal morphology with interconnecting processes and did not undergo rapid death. However, these cells did not differentiate further as judged from the continued absence of NF expression. Thus E-N-CAM+ neuronal progenitor cells isolated at different stages of brain development appear to have different microenvironmental requirements for differentiation and survival (see pages 364-370, under all sections entitled "Differentiation...", and page 370, right column, first two paragraphs).

Blass-Kampmann *et al.* do not disclose an initial culturing of cells obtained from the brain followed by isolating the neuron-restricted precursor cells, and a further culturing step. In addition, Blass-Kampmann *et al.* do not disclose the addition of chick embryo extract to the medium, or all of the claim-designated growth factors, differentiation factors, maturation factors, trophic factors, etc. However, Boss *et al.* disclose a method of culturing progenitor cells *in vitro* comprising using porcine or human embryos at Carnegie stages 15-18, dissociating cells from the ventral mesencephalon, and culturing the cells as monolayers. The cultures are initially grown in a first culture medium which promotes the survival of neuron progenitor cells which are capable of proliferating; the initial culture medium can be a basal medium supplemented with serum, hormones, growth factors, and trace elements. The cultures remain in the initial culture medium for at least 4.5 days prior to culture in growth medium (see, e.g., columns 6-8, under "Isolation of Tissue"). Selections of specific cell types can be performed using FACS, and magnetic bead antibody sorting (see, e.g., column 12, lines 52-58). Cells are seeded on poly-L-ornithine coated tissue culture plastic and are passaged prior to extensive neurite formation (see, e.g., column 11, lines 57-68, and column 12, lines 1-60). The neuron progenitor cells can be induced to differentiate *in vitro* by adding a differentiation agent to the culture medium. The neuron progenitor cells are switched from an initial medium to a growth medium, and 0.4, 2.0, or 10.0 mM dbc-AMP is added to the medium to induce differentiation (see, e.g., Example 8). The

cultured cells produced the catecholamines dopamine, epinephrine, and norepinephrine The cells can be induced to differentiate *in vitro* by replacing the growth medium with replacement medium containing the differentiation agent such as retinoic acid and di-butyryl cyclic AMP which results in differentiated cell which secrete dopamine, epinephrine, and norepinephrine (column 13, lines 58-60, and column 20, lines 41-64). Boss *et al.* also disclose a composition comprising neuron progenitor cells in a physiologically acceptable buffer (see, e.g., column 14, lines 10-25). Similarly, Johe *et al.* disclose dissociating cells from various regions of embryonic brain and culturing the cells in the presence of either EGF (20 ng/ml) or bFGF (10 ng/ml) to allow proliferation of the cells. TGF-a (10 ng/ml) was also a mitogen for the multipotential cells. EGF- and bFGF-expanded colonies were also differentiated by withdrawing the mitogens (see, e.g., column 16, lines 40-68). In addition, Johe *et al.* tested the influence of growth factors on cells of embryonic and adult rat brain by adding the growth factors to the culture two days prior to withdrawal of bFGF and during the 6 days of differentiation. Factors tested included PDGF-AA, -AB, or -BB, CNTF, T3, NGF, NT-3, BDNF, TGF-β1, IL1b, IL2-11, G-CSF, M-CSF, GM-CSF, oncostatin M, stem cell factor, erythropoietin, interferon gamma, 9-cis and all-trans retinoic acid, retinyl acetate, dexamethasone, and corticosterone (see, e.g., column 17, lines 54-59). Moreover, Weiss *et al.* disclose that cells which are derived from the neural tube give rise to neurons and glia of the central nervous system, and certain factors present during development, such as nerve growth factor, promote growth of neural cells (see page 2, lines 8-19). Cells can be obtained from donor tissue by tissue dissociation techniques and placed into any known culture medium capable of supporting cell growth. Physiological conditions are approximated for culturing of the cells. The cells can be grown in suspension or on a fixed substrate. Culturing of cells on a fixed substrate can result in differentiation of the progenitor into a terminally differentiated cell which is not longer capable of dividing, i.e., is postmitotic (see page 17, line 14, through page 19, line 10). Differentiation of cells can be induced by plating the cells on a fixed substrate including poly-L-lysine or poly-L-ornithine coated flasks, plates, or coverslips in the presence of EGF, TGFα or any factor capable of sustaining differentiation, such as basic fibroblast growth factor (see page

22, lines 3-12). Neuronal cells derived from the culture are identified using antibodies specific for known cellular markers, neurotransmitters, neurotransmitter-synthesizing enzymes, or neurotransmitter receptors (see page 22, lines 13, through page 23, line 24). Moreover, antibodies to an intermediate filament protein, found specifically in neuroepithelial stem cells can be used in cellular identification (see page 24, lines 1-8). Furthermore, Rao *et al.* (26th Annual Meeting of the Society for Neuroscience, 22:527, Abstract #215.12, 1996) disclose that cultures of E10.5 neuroepithelial cells from the rat caudal neural tube require both FGF and chick embryo extract to proliferate and to maintain an undifferentiated phenotype (see abstract), and Lee *et al.* disclose culturing NT2 cells on substrata such as Matrigel (which is known in the art to contain the claim-designated RGD-containing proteins), poly-D-lysine, or laminin (see, e .g, column 5, lines 34-37, and column 9, lines 41-45). The cells can be differentiated into greater than 95% pure cultures of neuronal cells when cultured in the presence of retinoic acid (see, e.g., column 4, lines 39-68), and that the neuronal cells can be maintained in a post-mitotic state after withdrawal of mitotic inhibitors (see, e.g., column 6, lines 26-35).

It would have been obvious to one of ordinary skill in the art at the time the claimed invention was made to modify the cell culture methods of Boss *et al.*, Weiss *et al.*, or Johe *et al.* by isolating specific populations of lineage restricted cells, such as those disclosed by Blass-Kampmann *et al.*, to determine the effect of different *in vivo* developmental stages on the ability of precursor cells to differentiate into various cell types of the central nervous system, and to determine the effect of various growth factors, such as fibroblast growth factor and chick embryo extract, as disclosed by Rao *et al.*, or different maturation factors, as disclosed by Boss *et al.*, Weiss *et al.*, or Johe *et al.*, or different substrata, as disclosed by Lee *et al.*, for directing the precursor cells into different lineages. In view of the combined teachings, one of ordinary skill in the art would have had a high expectation of successfully obtaining neuronal cells at a specific stage of differentiation by isolating the neuronal cells during the claim-designated stages of *in vivo* development, using an antibody which binds to a cell surface marker expressed during that particular stage of differentiation, such as embryonic neural cell adhesion molecule, prior to or

after culturing, using any of the claim-designated isolation protocols absent evidence to the contrary. Thus, from the combined teachings, it would have been obvious to one of ordinary skill in the art to isolate a population of neuronal precursors, expand the precursors *in vitro* under the appropriate cell culture conditions, and differentiate the cells into neurons as these cells can be utilized to study various aspects of neural cell biology.

Thus the claimed invention as a whole was clearly *prima facie* obvious at the time the claimed invention was made especially in the absence of sufficient, clear, and convincing evidence to the contrary.

***Double Patenting***

A rejection based on double patenting of the "same invention" type finds its support in the language of 35 U.S.C. 101 which states that "whoever invents or discovers any new and useful process ... may obtain a patent therefor ..." (Emphasis added). Thus, the term "same invention," in this context, means an invention drawn to identical subject matter. See *Miller v. Eagle Mfg. Co.*, 151 U.S. 186 (1894); *In re Ockert*, 245 F.2d 467, 114 USPQ 330 (CCPA 1957); and *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970).

A statutory type (35 U.S.C. 101) double patenting rejection can be overcome by canceling or amending the conflicting claims so they are no longer coextensive in scope. The filing of a terminal disclaimer cannot overcome a double patenting rejection based upon 35 U.S.C. 101.

Claims 1-7, 12-16, 28-32, of this application conflict with claims 1-12, and 24-26 of Application No. 08/909,435. 37 CFR 1.78(b) provides that when two or more applications filed by the same applicant contain conflicting claims, elimination of such claims from all but one application may be required in the absence of good and sufficient reason for their retention during pendency in more than one application. Applicant is required to either cancel the conflicting claims from all but one application or maintain a clear line of demarcation between the applications. See MPEP § 822.

The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. See *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Ornum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970);and, *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) may be used to overcome an actual or provisional rejection based on a nonstatutory double patenting ground provided the conflicting application or patent is shown to be commonly owned with this application. See 37 CFR 1.130(b).

Effective January 1, 1994, a registered attorney or agent of record may sign a terminal disclaimer. A terminal disclaimer signed by the assignee must fully comply with 37 CFR 3.73(b).

Claims 8-11, 17-29, 33, and 34 are provisionally rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 13-23 and 27-29 of copending Application No. 08/909,435. Although the conflicting claims are not identical, they are not patentably distinct from each other because modifications in the methods of isolating the neuron-restricted progenitor cells, including the source of the tissue or cell population, addition of different growth and/or maturation factors, and obtaining differentiated cells with the claim-designated neuronal characteristics would have been obvious modifications and well within the purview of the skilled artisan.

This is a provisional obviousness-type double patenting rejection because the conflicting claims have not in fact been patented.

***Sequence Compliance***

This application contains sequence disclosures that are encompassed by the definitions for nucleotide and/or amino acid sequences set forth in 37 CFR 1.821(a)(1) and (a)(2). However, this application fails to comply with the requirements of 37 CFR 1.821 through 1.825 for the reason(s) set forth on the attached Notice To Comply With Requirements For Patent Applications

Containing Nucleotide Sequence And/Or Amino Acid Sequence Disclosures. Applicant is requested to return a copy of the attached Notice to Comply with the response.

No claims are allowed.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Janet M. Kerr whose telephone number is (703) 305-4055. Should the examiner be unavailable, inquiries should be directed to Brian Stanton, Supervisory Primary Examiner of Art Unit 1633, at (703) 308-2801. Papers related to this application may be submitted to Group 1600 by facsimile transmission. Papers should be faxed to Group 1600 via the PTO Fax Center located in Crystal Mall 1. The faxing of such papers must conform with the notice published in the Official Gazette, 1096 OG 30 (November 15, 1989). The CM1 Fax Center number is (703) 305-7401. Any inquiry of a general nature or relating to the status of this application should be directed to the Group 1600 receptionist whose telephone number is (703) 308-0196.

The Group and/or Art Unit location of your application in the PTO has changed. To aid in correlating any papers for this application, all further correspondence regarding this application should be directed to Group Art Unit 1633.

Janet M. Kerr, Ph.D.
Patent Examiner
Group 1600

Karen M. Hauda
Karen M. Hauda
Patent Examiner